

## ***SDBI-1900, SMO-015 - Integrated Immune***

### **Validation of Procedures for Monitoring Crewmember Immune Function**

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**Background:** The objective of this Supplemental Medical Objective (SMO) is to determine the status of the immune system, physiological stress and latent viral reactivation (a clinical outcome that can be measured) during both short and long-duration spaceflight. In addition, this study will develop and validate an immune monitoring strategy consistent with operational flight requirements and constraints. Pre-mission, in-flight and post-flight blood and saliva samples will be obtained from participating crewmembers. Assays included peripheral immunophenotype, T cell function, cytokine profiles, viral-specific immunity, latent viral reactivation (EBV, CMV, VZV), and stress hormone measurements. To date, 18 short duration (now completed) and 8 long-duration crewmembers have completed the study. The long-duration phase of this study is ongoing. For this presentation, the final data set for the short duration subjects will be discussed.

**General Immune Status:** The constitutive distribution of most peripheral leukocyte subset populations is largely unaltered during flight. Exceptions include a mild increase in levels of memory CD4+ T cells, and a decrease in naïve CD8+ T cells accompanied by a corresponding increase in central/effector memory CD8+ T cells. No increase in constitutively activated T cells was observed during flight. Various functional measurements were employed. Cytokine producing T cells (intracellular measurement; both CD4+/IL-2+ and CD8+/IFN $\gamma$ +) were mildly reduced during flight and further reduced upon landing. General T cell function (early blastogenesis response to mitogenic stimulation), yielded varying results. T cell stimulation with Staphylococcal enterotoxins was dramatically reduced in-flight, whereas T cell stimulation with anti-TCR antibodies was unchanged in-flight (and elevated post-flight). The post-flight elevation is in concurrence with previously published findings for that mitogen. Bulk secreted Th1/Th2 cytokines were measured following T cell activation, and mitogen-dependant in-flight reductions were observed in IFN $\gamma$ , IL-10, TNF $\alpha$  and IL-6 production. Secreted inflammatory cytokines levels were also measured following monocyte stimulation (LPS), however a flight-associated decrease was only observed for IL-10, whereas IL-8 levels were increased during flight.

**Viral Specific Immunity:** The number of virus-specific CD8+ T-cells was measured using MHC tetramers, while their function was measured using intracellular cytokine analysis following peptide stimulation. Both the number and function of EBV-specific cells decreased during flight as compared to preflight levels. The number of CMV-specific T-cells generally increased as the mission progressed while their function was generally unaltered. Viral (EBV) load in blood was elevated postflight. Elevated anti-EBV VCA antibodies were evident in ~40% of the astronauts; anti-CMV antibodies generally increased during and after flight.

**Latent viral reactivation:** Samples collected from crewmembers before, during and after the space flights were analyzed by the real time polymerase chain reaction (PCR) for the presence of virus (saliva for Epstein-Barr virus (EBV), Varicella zoster virus (VZV), and urine for cytomegalovirus (CMV) DNA. Higher levels of salivary EBV were found in during the flight-phase than before and after the flight as well as than the healthy control subjects. VZV was detected in about 50 % of the astronauts during and up to 5 days after space flight. No VZV was found in any preflight or control samples. There was also no CMV detected in any of the urine samples collected from astronauts or from healthy controls. However, CMV was shed in 35 % the inflight samples and 30% of postflight urine samples of the crewmembers.

**Physiological Stress:** Stress hormones were measured in plasma, urine and saliva before, during and after the spaceflight. There was generally a higher level of cortisol as measured in blood, urine and saliva in the

astronauts during flight. Circadian rhythm of salivary cortisol was normal before and after flight in most of the astronauts, however, changes were observed during the flight phase.